

3406

1/18

AAGCTTAAGGAGGTTAATCG ATG AAA AAA ACT GCT ATC GCT ATC GCT GTT

H A C met

i f RBS

n l a

3 2 1

GCT CTG GCT GGT TTC GCT ACT GTT GCT CAG GCG GCG CCG AGA TCT

ala N

a r

1 2

AAA CAG GAA TTC GAG CTC GGT ACC CGG GGA TCC TCT AGA GTC GAC

E S K X B X S

C a p m a b a l

O c h a m a l

I 1 1 1 1 1 1

CTG CAG GCA TGC

P S

s p

t h

1 1

3557

cI857

P_R

RBS

7 NUCLEOTIDES

ClaI

NarI

MULTIPLE CLONING SEQUENCE

NarI-SphI

SphI

rno-INDEPENDENT
TRANSCRIPTIONS
TERMINATING
SEQUENCES

pHD389

(4286 bp)

pUC19

ori

SIGNAL PEPTIDE FOR THE
SEQUENCE FROM ompARBS = RIBOSOMAL
BINDING SEQUENCEP_R = "RIGHT" PROMOTOR FROM COLIPHAGE λcI857 THE GENE FOR A HEAT-SENSITIVE REPRES-
SOR-PROTEIN FROM COLIPHAGE λ

FIG. 1 PLASMA pHD 389. THE RIBOSOMAL
BINDING-SEQUENCE (EMPHASIZED WITH
A FULL LINE), THE SEQUENCE FOR SIGNAL PEPTIDE FROM ompA
(FROM E.coli) (DOTTED LINE) AND RECOGNITION SEQUENCE FOR
SEVERAL RESTRICTION ENZYMES ARE SHOWN.

MULTIPLE CLONING SEQUENCE

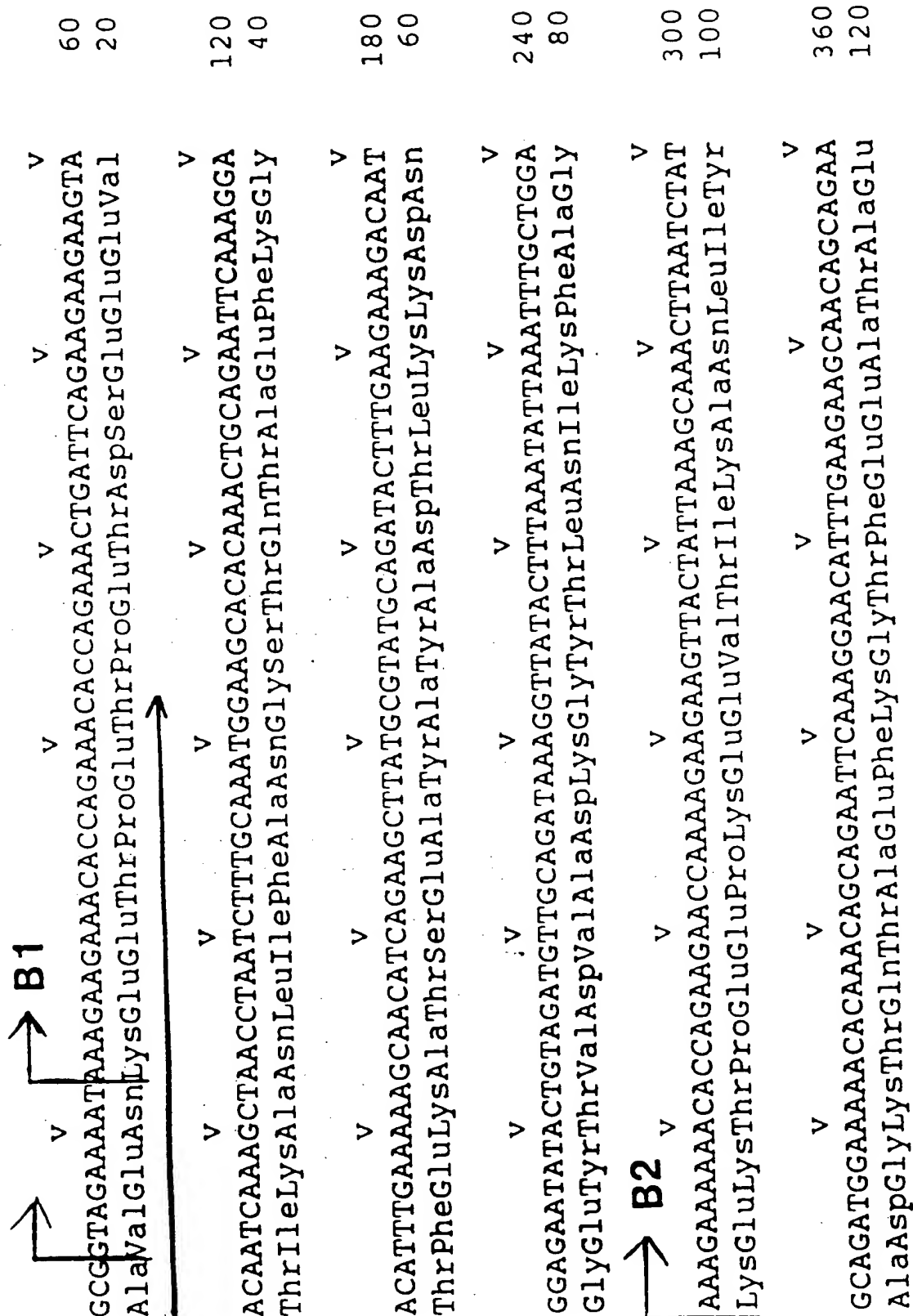
NarI
BglII
EcoRI
SacI
KpnI
XmaI
BamHI
XbaI
SalI
PstI
SphI

UNIQUE
CLEAVING
SITES

08/325278

2/18

PROTEIN LG

FIG. 2
(CONT.)

3/18

GCATACAGATATGCAGATGCAATTAAGAAGGACAAATGGAGAAATATACAGTAGACGTTGCA 420
AlaTyrArgTyrAlaAspAlaLeuLysLysAspAsnGlyGluTyrThrValAspValAla 140

GATAAAGGTTACTTTAAATATTAATTTGCTGGAAAAGAAAAAACACACAGAAACCA 480
AspLysGlyTyrThrLeuAsnIleLysPheAlaGlyLysGluLysThrProGluGluPro 160

AAAGAAGAAGTTACTATTAAGCAAACTTAATCTATGCAGATGGAAAAACACAAACAGCA 540
LysGluGluValThrIleLysAlaAsnLeuIleTyrAlaAspGlyLysThrGlnThrAla 180

GAATTCAAAGGAACATTTGAAGAAGCAACAGCAGCAAGCATACAGATATGCTGACTTATTA 600
GluPheLysGlyThrPheGluGluAlaThrAlaGluAlaTyrArgTyrAlaAspLeuLeu 200

GCAAAGAAATCGTAAATATACAGTAGACGTTGCAGATAAAGGTTATACCTTTAAATATT 660
AlaLysGluAsnGlyLysTyrThrValaspValAlaAspLysGlyTyrThrLeuAsnIle 220

AAATTGCTGGA^{B4}AAAGAAAAACACCAGAAGAACCAAAAGAAAGTACTATTAAAGCA 720
LysPheAlaGlyLysGluLysThrProGluGluProLysGluGluValThrIleLysAla 240

AACTTAATCTATGCAGATGGAAAAA^{B3}CTCAAACAGCAGAGTTCAAAGGAACATTTGCAGAA 780
AsnLeuIleTyrAlaAspGlyLysThrGlnThrAlaGluPheLysGlyThrPheAlaGlu 260

FIG. 2
(CONT.)

4 / 18

FIG. 2
(CONT.)

5/18

V V V V V V V
 AAACCTGTTAATGGTAAACATTTGAAAGCGCAACTACTAAAGCAGTAGACGCA 1200
 LysLeuValIleAsnGlyLysThrLeuLysGlyGluThrThrThrLysAlaValAspAla 400

V V V V V V V
 GAAACTGCAGAAAAGCCCTTCAAAACAATACGCTAACGACAACGGTGTGATGGTGTGG 1260
 GluThrAlaGluLysAlaPheLysGlnTyrAlaAsnAspAsnGlyValAspGlyValTrp 420

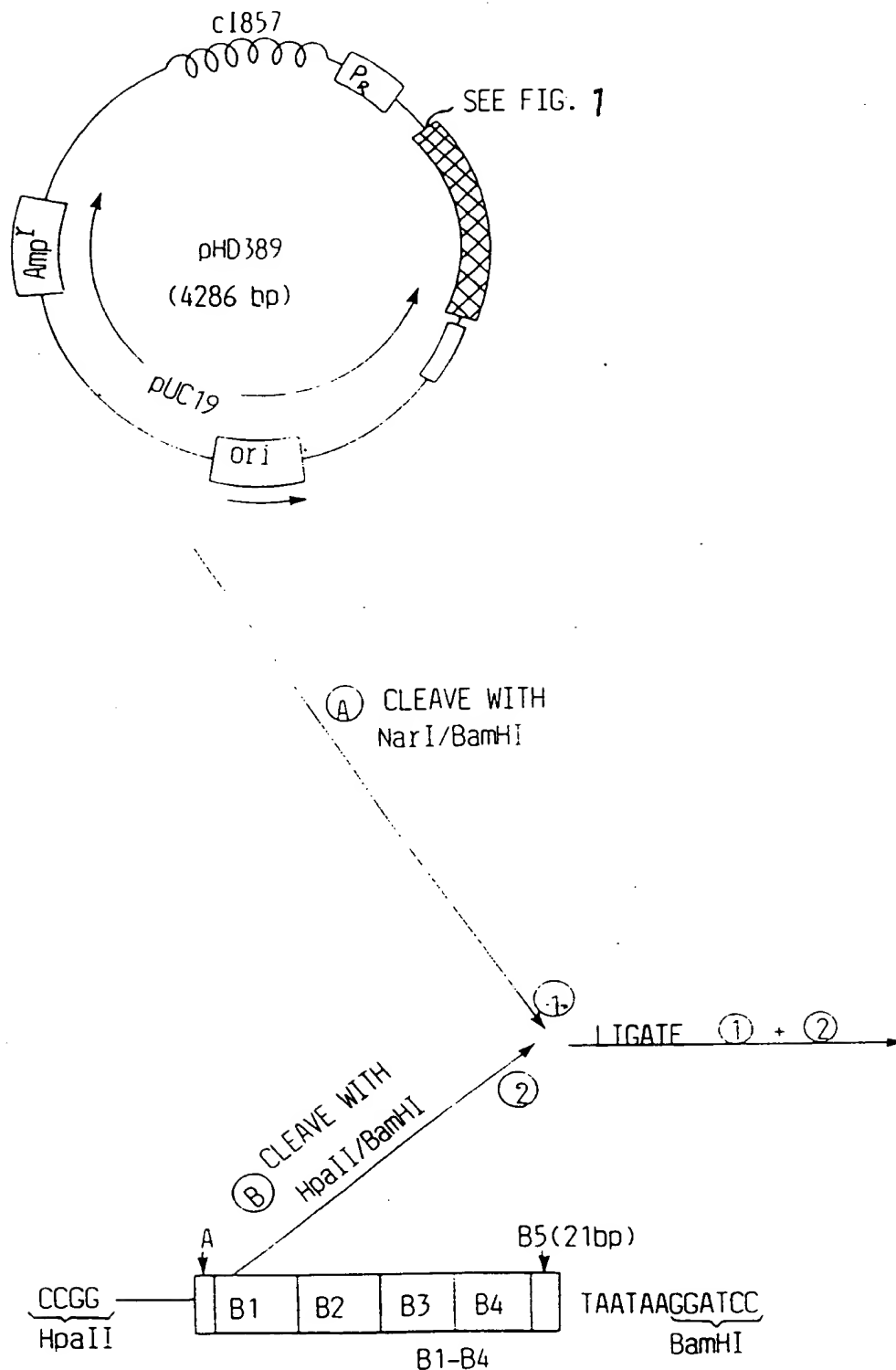
V V V V V
 ACTTATGATGCGACTAAGACCTTTACGGTAACTGAAATGTAATAA 1308
 ThrTyrAspAspAlaThrLysThrPheThrValThrGluMet - - 434

FIG. 2

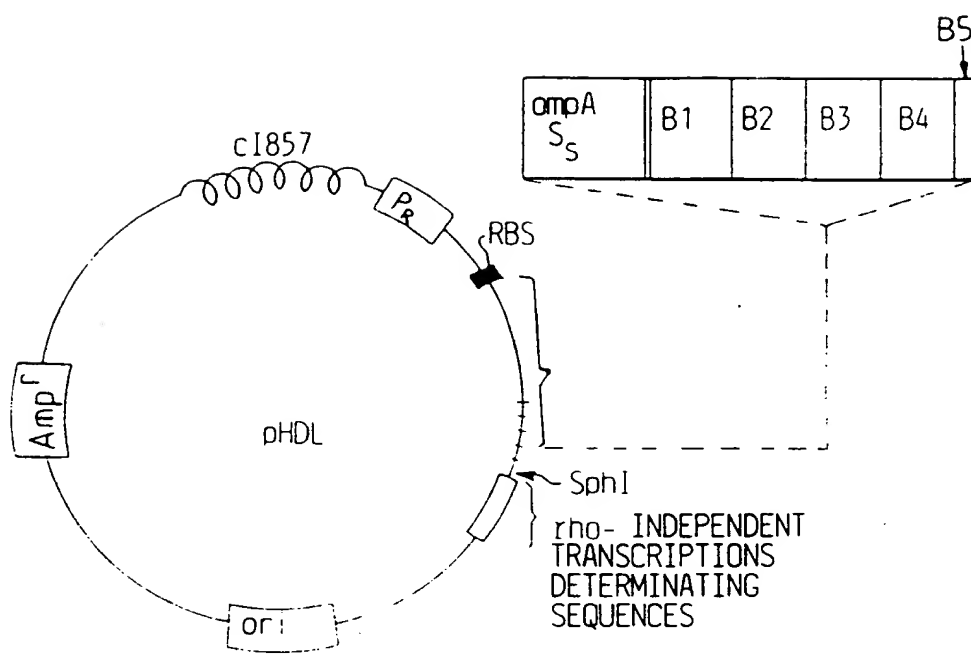


6/18

FIG. 3 SCHEMATIC OVERALL VIEW OF THE PRODUCTION OF PROTEIN L



7/18



TRANSFORM
TO E. coli
STRAIN LE392

FIG.3(2)

8/18

FIG. 4

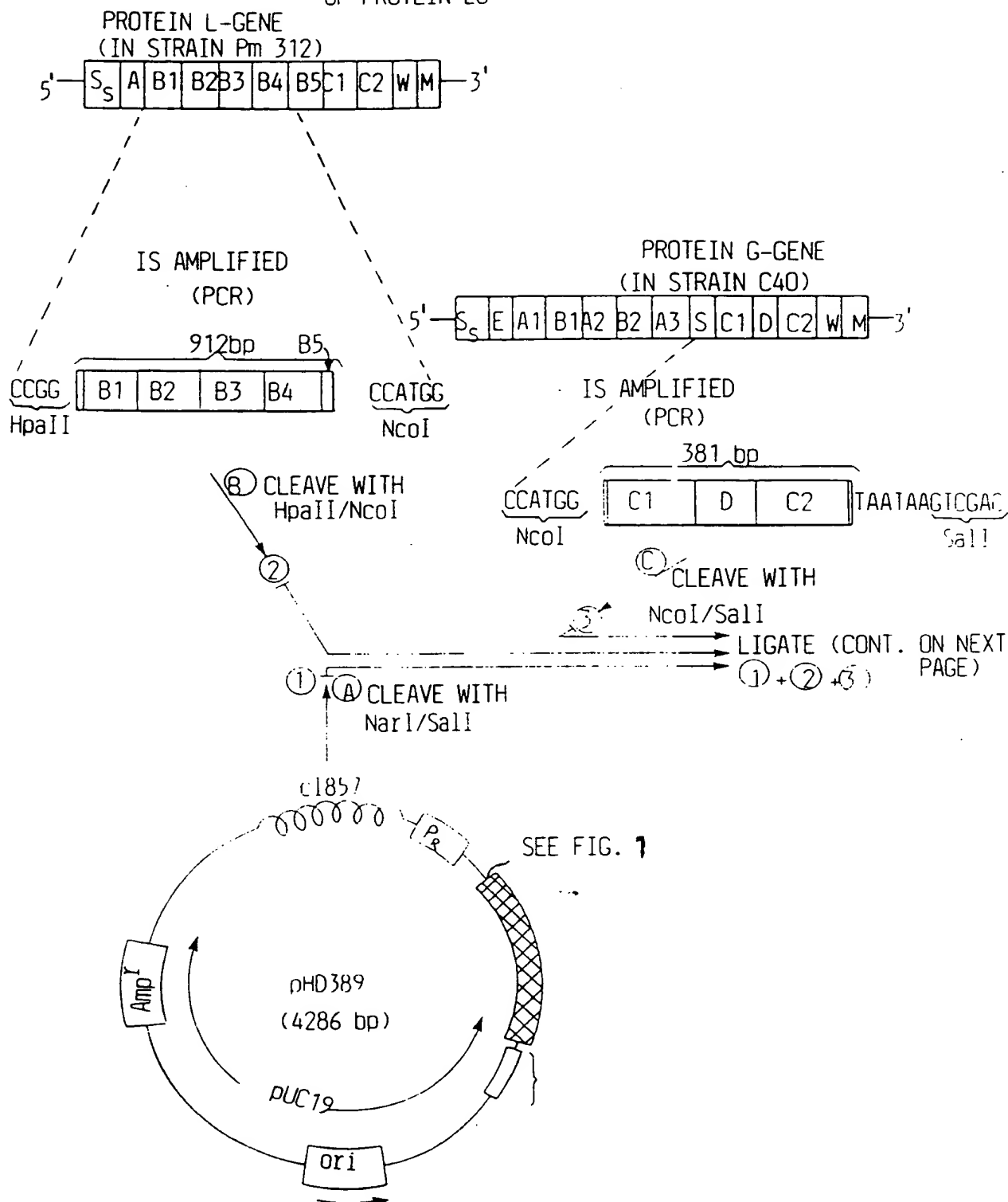
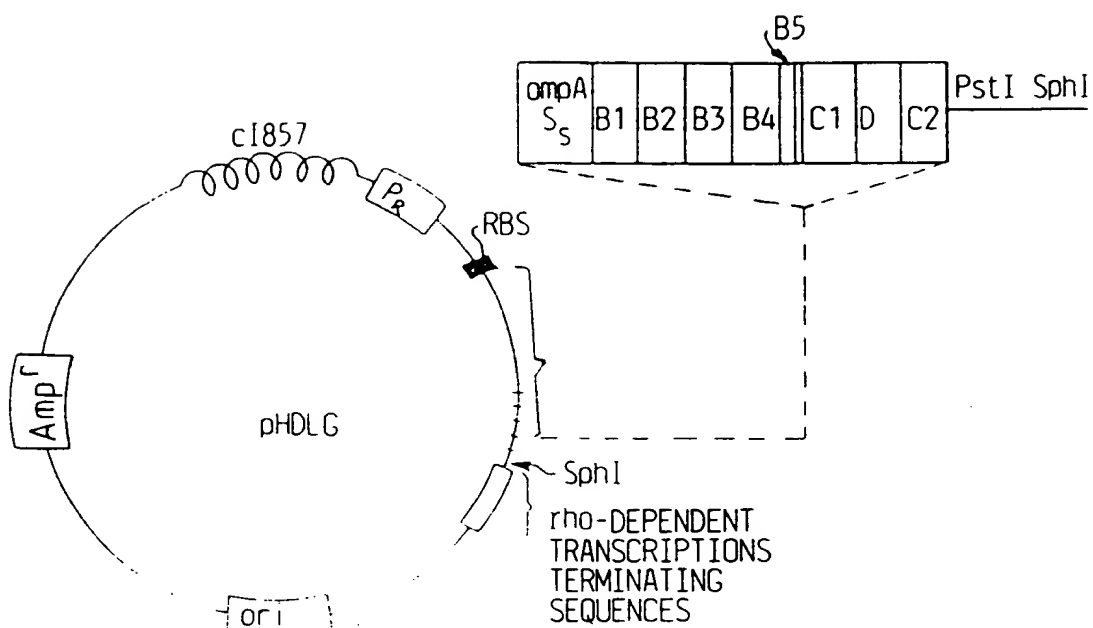
SCHEMATIC OVERALL VIEW OF PRODUCTION
OF PROTEIN LG

FIG.4(1)

9/18

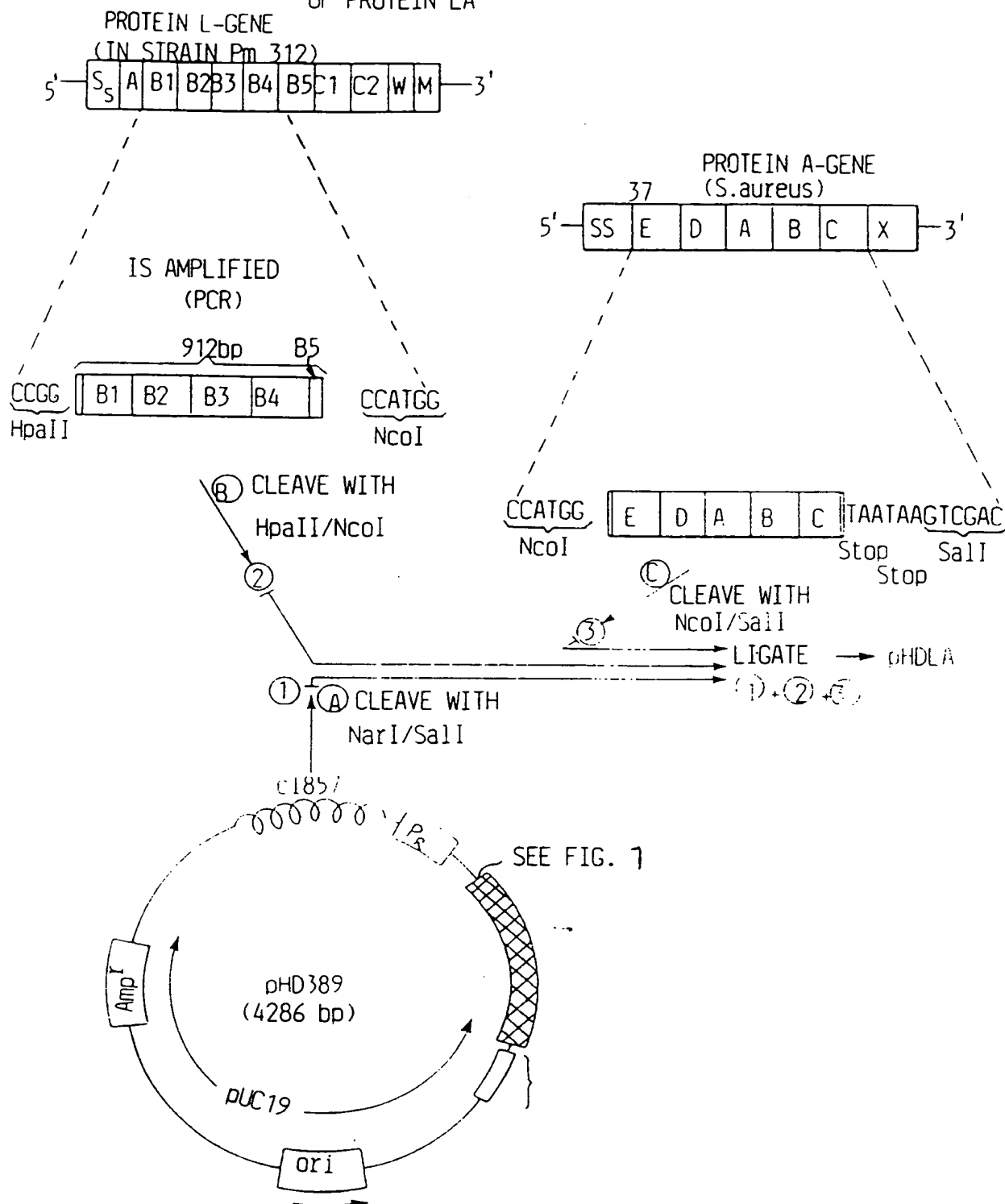


TRANSFORM
TO *E. coli*
STRAIN LE392

FIG.4(2)

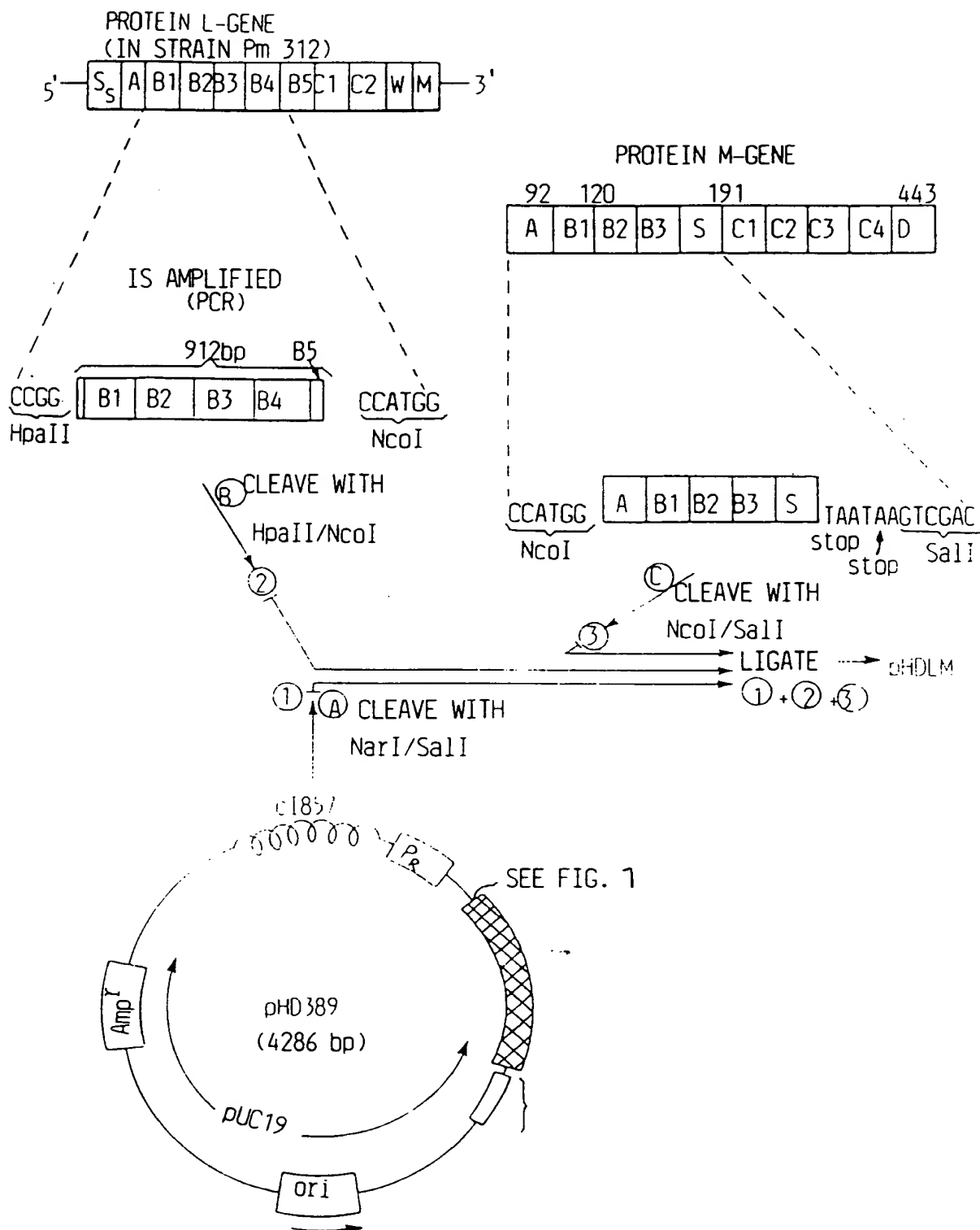
10/18

FIG. 5a

SCHEMATIC OVERALL VIEW OF THE PRODUCTION
OF PROTEIN LA

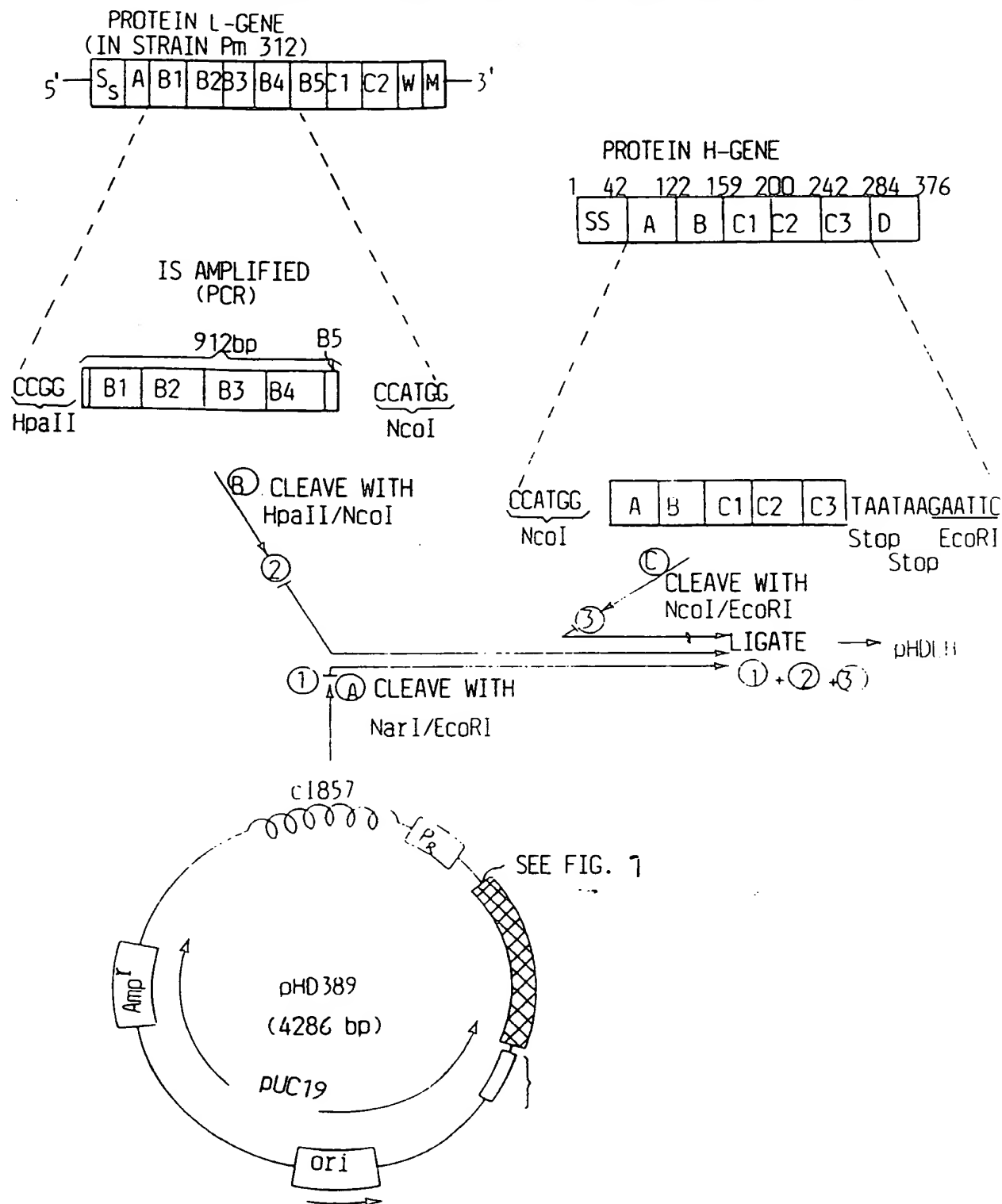
11/18

FIG. 5b SCHEMATIC OVERALL VIEW OF THE PRODUCTION OF PROTEIN LM



12/18

FIG. 5c SCHEMATIC OVERALL VIEW OF THE PRODUCTION OF PROTEIN LH





14/18

AACGGTGTATCCTAGGGAAGTTATAGAGATCTTGCAGCAACAAATCCCGCAATA
 AsnGlyAspGlyAsnProArgGluValIleGluAspLeuAlaIleAsnAsnProAlaIle
 60 20
 CAAAATATACGTTTACGTCACGAAACACAGGACTTAAAGCGGAGATTAGAGAATGCAATG
 GlnAsnIleArgLeuArgHisGluAsnLysAspLeuLysAlaArgLeuGluAsnAlaMet
 120 40
 GAAGTTGCAGGAAGAGATTTTAAAGAGAGCTGAAGAACTTGAAAGCAAAACAGCCTTA
 GluValAlaGlyArgAspPheLysArgAlaGluGluLeuGluLysAlaLysGlnAlaLeu
 180 60
 GAAGACCAGCGTAAAGATTTAGAACTAAATTAAAGAACTACACACAGACTATGACTTA
 GluAspGlnArgLysAspLeuGluThrLysLeuLysGluLeuGlnGlnAspTyrAspLeu
 240 80
 GCAAAGGAATCAACAAGTTGGGATAGACAAAGACTTGAAAGAGGTTAGAGAGAGAAAAG
 AlaLysGluSerThrSerTrpAspArgGlnArgLeuGluLysGluLeuGluGluLysLys
 300 100
 GAAGCTCTTGAAATTAGCGATAGACCAGGCAAGTCGGGACTACCATAGAGCTACCGCTTTA
 GluAlaLeuGluLeuAlaIleAspGlnAlaSerArgAspTyrHisArgAlaThrAlaLeu
 360 120
 GAAAAAGAGTTAGAGAGAGAAAGAAAGCTCTTGAAATTAGCGATAGACCAAGCGAGTCAG
 GluLysGluLeuGluGluLysLysLysAlaLeuGluLeuAlaIleAspGlnAlaSerGln
 420 140
 GACTATAATAGAGCTAACGCTCTTAGAAAAAGAGTTAGAAACGATTACTAGAGAACAGAG
 AspTyrAsnArgAlaAsnValLeuGluLysGluLeuGluThrIleThrArgGluGlnGlu
 480 160
 ATTAATCGTAATCTTTTAGGCAATGCAAACTTGAACTTGATCAACTTTCATCTGAAAAA
 IleAsnArgAsnLeuLeuGluGlyAsnAlaLysLeuGluLeuAspGlnLeuSerSerGluLys
 540 180

FIG. 7 (CONT.)

GAGCAGCTAACGATCGAAAGCAAACTT^VBAGGAAGAAACAAATCTCAGACGCAAGT^V 600
 GluGlnLeuThrIleGluLysAlaLysLeu^V190GluGluLysGlnIleSerAspAlaSer^V 200
 CGTCAAAGCCTTCGTGCTGACTTGGACGCATCAGCTAAGCTAAGAAACAGGTTGAAGAA^V 660
 ArgGlnSerLeuArgArgAspLeuAspAlaSerArgGluAlaLysLysGlnValGluLys^V 220
 GATTTAGCAAACTTGACTGCTGAACCTTGATAAGGTTAAAGAGAGACAAACAATCTCAGAC^V 720
 AspLeuAlaAsnLeuThrAlaGluLeuAspLysValLysGluAspLysGlnIleSerAsp^V 240
 GCAAGCCGTCAACGGCTTCGCCGTGACTTGGACGCATCAGCTAAGCTAAGAAACAGGTT^V 780
 AlaSerArgGlnArgLeuArgArgAspLeuAspAlaSerArgGluAlaLysLys^V

15/18

FIG.7 (CONT.)

Amino acid sequence and nucleic acid sequence for protein M1, IgG-binding somewhere between amino acid 1-190.

16/18

GAAAAAGATTAGCAAACTTGACTGCTGACTTGACTTGATAAGGTTAAAGAGAGAAAAACAATC 840
 GluLysAspLeuAlaAsnLeuThrAlaGluLeuAspLysValLysGluLysGlnIle 290
 TCAGACGCAAGCCGTCACGGCTTCGCCGTGACTTGGACGCATCACGCTGAGCTAAGAA 900
 SerAspAlaSerArgGlnArgLeuArgArgAspLeuAspAlaSerArgGluAlaLysLys 300
 CAAGTTGAAAAAGCTTTAGAAGAGCAACACAGCAAAATTAGCTGCTCTTGAAAAAATTAAAC 960
 GlnValGluLysAlaLeuGluGluAlaAsnSerLysLeuAlaAlaLeuGluLysLeuAsn 320
 AAAGAGCTTGAAAGAAAGCAAGAAATTAAACAGAAAAAGAAAGCTGAACCTACAAGCAAAA 1020
 LysGluLeuGluSerLysLysLeuThrGluLysGluLysAlaGluLeuGlnAlaLys 340
 CTTGAAGCAGAAAGCAAAAGCACTCAAGAAACAATTAGCGAAACAAGCTGAAGAACTCGCA 1080
 LeuGluAlaGluAlaLysAlaLeuLysGluGlnLeuAlaLysGlnAlaGluLeuAla 360
 AAATAAGAGCTGGAAAAAGCATCAGACTCACAAACCCCTGATACAAACCAAGCAACCAAA 1140
 LysLeuArgAlaGluLysAlaSerAspSerGlnThrProAspThrLysProGluAsnLys 380
 GCTGTTCCAGGTAAAGGTCAAGCACCACAGCAGGTACAAACCTAACCAAAACAAGCA 1200
 AlaValProGluLysGluGlnAlaProGlnAlaGluThrLysProAsnGlnAsnLysAla 400
 CCAATGAAGGAACCTAAGAGACAGTTACCATCAACAGGTGAACAGCTAACCCATTCTTC 1260
 ProMetLysGluThrLysArgGlnLeuProSerThrGluGluThrAlaAsnProPhe 420
 ACAGCGGCACGCGTTACTGTTATGGCAACAGCTGGAGTAGCAGCAGTTGTAAACGCAAA 1320
 ThrAlaAlaArgValThrValMetAlaThrAlaGluValAlaAlaValValLysArgLys 440

FIG. 7

GAAGAAAACTAA 1329
 GluGluAsn >>> 443

18/18

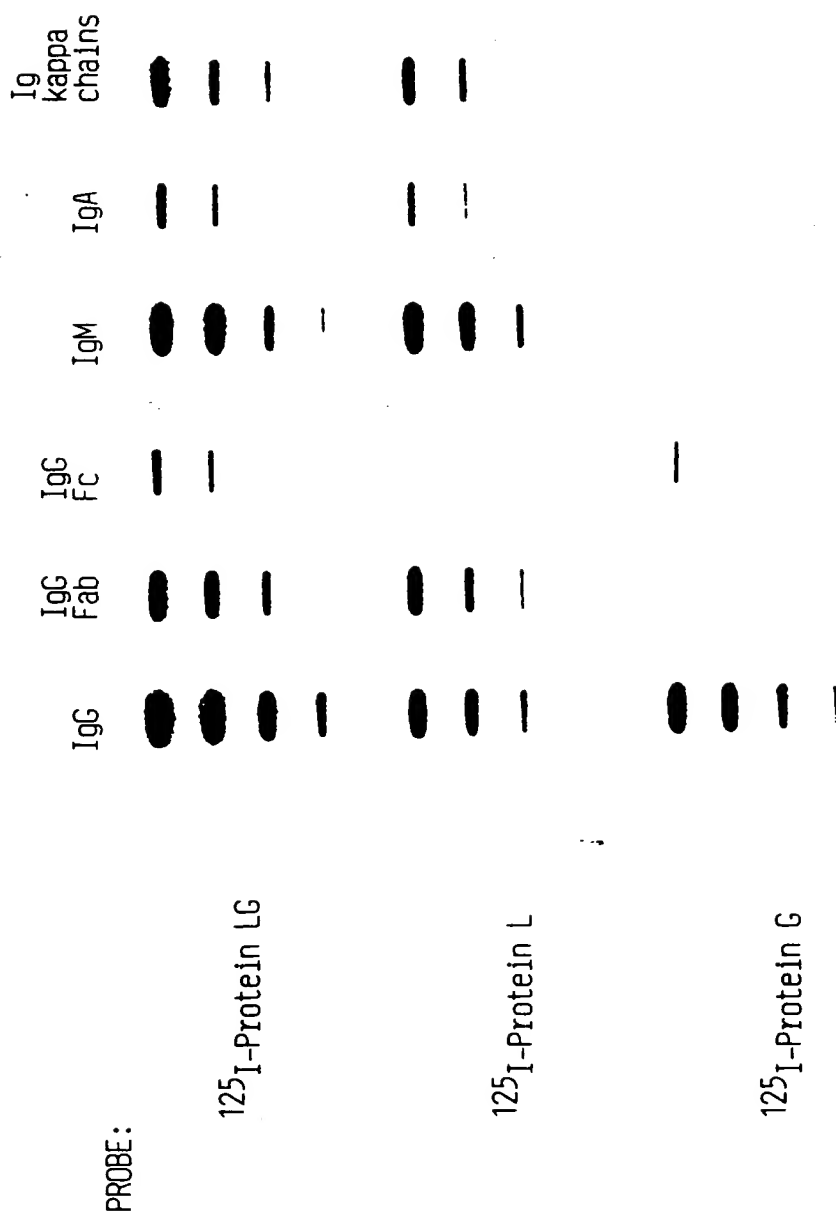


FIG. 9